4

Hazard Characterization

This chapter describes the process used to characterize the number of symptomatic infections resulting from the consumption of cooked ground beef servings contaminated with *Escherichia coli* O157:H7. This process is commonly referred to as a dose-response assessment. Unlike many chemical hazards and some pathogens, the dose-response relationship for *E. coli* O157:H7 is unknown. Limited dose-response information is available from an animal study conducted by Pai et al. (1986), in which infant rabbits were exposed to *E. coli* O157:H7. Because there is no effective treatment for *E. coli* O157:H7 infection and the outcome of infection can include severe illness and death, experimental studies exposing humans to *E. coli* O157:H7 have not been, and probably never will be, performed. In contrast, a substantial amount of surveillance data exist on the annual number of illnesses due to infection with *E. coli* O157:H7. Thus, this risk assessment uses a multistep process to derive a dose-response function for E. *coli* O157:H7 (Figure 4-1). This process is divided into four primary steps: (1) estimation of the number of *E. coli* O157:H7-related illnesses attributable to the consumption of contaminated ground beef (response); (2) estimation of the likelihood and level of *E. coli* O157:H7 in cooked ground beef servings (dose, derived in Chapter 3); (3) derivation of upper and lower bounds for the *E. coli* O157:H7 dose-response function based on clinical studies of surrogate pathogens; and (4) derivation of the "most likely" (50th percentile) dose-response function for *E. coli* O157:H7 (Powell et al. 2000).

This chapter begins by estimating a baseline annual number of illnesses of *E. coli* O157:H7 infection from all exposures using data from the Emerging Infections Program, Foodborne Disease Active Surveillance Network (FoodNet). This baseline annual number of cases is adjusted upward to account for underdiagnosis and underreporting, providing an estimated total annual number of cases of symptomatic *E. coli* O157:H7 infection for the United States. Then, using data from studies of sporadic cases of *E. coli* O157:H7 infection and outbreaks of *E. coli* O157:H7, the proportion of total cases due to ground beef exposure is derived. Next, lower and upper bound dose-response functions are constructed using foodborne pathogens other than *E coli* O157:H7. These lower and upper bound dose response functions are used in combination with the estimated number of cases due to ground beef and the estimated number of ground beef

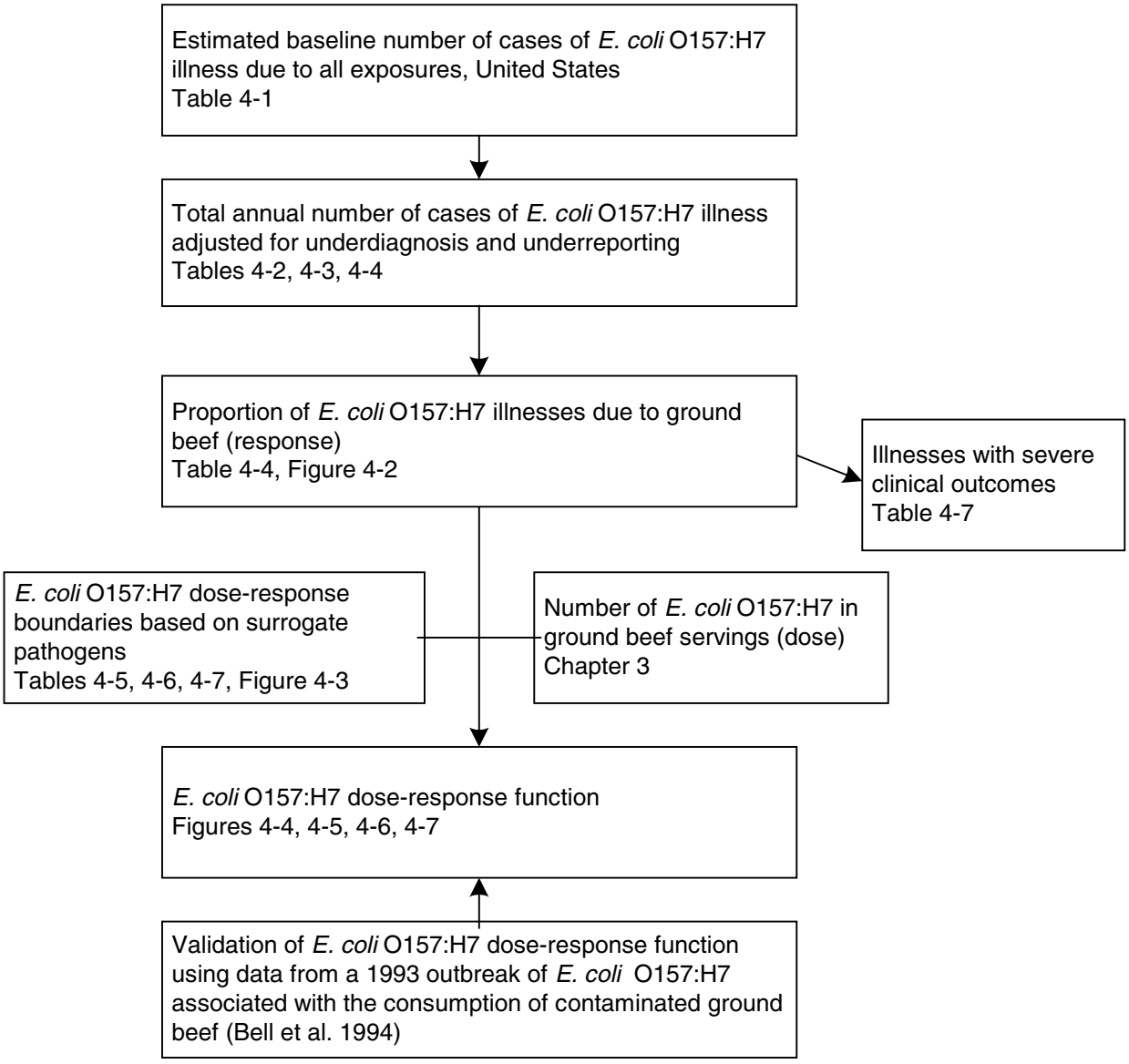

FIGURE 4-1 Flowchart for the derivation of the dose-response function for *E. coli* O157:H7 in ground beef.

servings contaminated with *E. coli* O157:H7 to generate a dose-response function for *E. coli* O157:H7. Finally, dose and response information from an outbreak of *E. coli* O157:H7 due to contaminated ground beef is compared with the *E. coli* O157:H7 dose-response function.

DEFINITION OF KEY TERMS

The following key terms are used throughout this chapter:

- Dose is the number of *E. coli* O157:H7 organisms in a serving of ground beef.
- Response refers to the number and severity of illnesses resulting from consumption of ground beef servings contaminated with *E. coli* O157:H7.

- Dose-response function refers to the mathematical relationship between the consumption of a ground beef serving containing a specific number (dose) of organisms and the resulting number of illnesses (response).
- Surrogate pathogens refers to pathogens that are either closely related genetically or have a similar mechanism of pathogenicity to the pathogen of interest.
- Lower bound refers to the dose-response curve derived from pathogenesis studies of enteropathogenic *E. coli* (EPEC).
- Upper bound refers to the dose-response curve derived from pathogenesis studies of *Shigella dysenteriae*.

ESTIMATING THE RESPONSE

Estimating the number of symptomatic *E. coli* O157:H7 infections due to contaminated ground beef first requires an estimation of the total number of cases that occur annually in the United States from all causes.

Baseline Annual Number of *E. coli* O157:H7 Infections Due to All Causes

FoodNet surveillance data for 1996 to 1999 were used to estimate the annual baseline number of symptomatic *E. coli* O157:H7 infections (1999 is the most recent year for which a final report is available). For information about FoodNet, please see Chapter 2. For each year and FoodNet site, the number of cases per 100,000 population was calculated. This rate per 100,000 population was multiplied by that state's population and then divided by the total population of all sites, for that year, providing a weighted rate for each state. Weighted rates for each state were then summed, resulting in an annual incidence estimate for 1996 to 1999 of 1.53, 1.25, 1.95, and 2.09, respectively (Table 4-1).

TABLE 4-1 Population-Weighted Rate of Illness Caused by *E. coli* O157:H7

	Year			
	1999	1998	1997	1996
FoodNet State: California				
Cases reported to FoodNet[a]	23	35	19	22
FoodNet catchment population[a]	2,162,359	2,063,454	2,063,454	2,063,454
Unadjusted rate (per 100,000 person-years)	1.06	1.70	0.92	1.07
State population[b]	33,145,121	32,666,550	32,182,118	31,762,190
Weighted rate	0.47	1.07	0.58	0.67
State: Connecticut				
Cases	94	58	34	38
Catchment population	3,282,031	2,460,127	2,460,127	1,626,366
Unadjusted rate	2.86	2.36	1.38	2.34
State population	3,282,031	3,274,069	3,267,240	3,263,910
Weighted rate	0.12	0.15	0.09	0.15

(continued)

TABLE 4-1 (continued)

	Year			
	1999	1998	1997	1996
State: Georgia				
Cases	44	51	8	15
Catchment population	7,788,240	3,541,230	3,541,230	2,729,783
Unadjusted rate	0.56	1.44	0.23	0.55
State population	7,788,240	7,642,207	7,489,982	7,334,183
Weighted rate	0.06	0.21	0.03	0.08
State: Maryland[c]				
Cases	16	24		
Catchment population	2,450,566	2,444,280		
Unadjusted rate	0.65	0.98		
State population	5,171,634	5,130,072		
Weighted rate	0.04	0.07		
State: Minnesota				
Cases	175	209	199	239
Catchment population	4,775,508	4,725,419	4,687,408	4,657,758
Unadjusted rate	0.65	4.42	4.25	5.13
State population	5,171,634	4,725,419	4,687,408	4,648,081
Weighted rate	0.04	0.41	0.39	0.47
State: New York[c]				
Cases	94	22		
Catchment population	20,844,453	1,106,085		
Unadjusted rate	4.51	1.99		
State population	18,196,601	18,159,175		
Weighted rate	1.08	0.48		
FoodNet State: Oregon				
Cases	64	101	80	73
Catchment population	3,316,154	3,281,974	3,243,272	3,203,735
Unadjusted rate	1.93	3.08	2.47	2.28
State population	3,316,154	3,281,974	3,243,272	3,195,409
Weighted rate	0.08	0.20	0.16	0.14
TOTAL				
Cases	510	500	340	387
Catchment population	25,859,311	19,622,569	15,995,491	14,281,096
Unadjusted rate	1.97	2.55	2.13	2.71
States' population	75,675,289	74,879,466	50,870,020	50,203,773
Weighted rate	2.09	1.95	1.25	1.53

[a]Data obtained from FoodNet final reports for each calendar year from www.cdc.gov.

[b]State population estimates for July 1 of each calendar year from www.census.gov.

[c]Maryland and New York began surveillance in 1998.

To represent variability in the annual number of reported cases, these four weighted rates were placed into a discrete uniform probability distribution (DUniform [1.53, 1.25, 1.95, 2.09]). During Monte Carlo simulation, one of these four rates is selected at random with equal probability. The output of this simulation is a list of possible rates and the frequency at which each possible rate occurred during multiple iterations of the model. The median rate from this output was multiplied by the estimated 1999 U.S. population of 272.7 million (Table 4-3) to obtain an estimated baseline annual number of cases of symptomatic *E. coli* O157:H7 infection. The median baseline number of cases estimated by the model was 4,200 (3,500 and 5,700—2.5th and 97.5th percentiles, respectively). This estimated number of cases has been rounded to two significant digits.

Adjusting the Baseline for Underdiagnosis and Underreporting

The baseline annual number of *E. coli* O157:H7 cases was adjusted upward to account for recognized sources of underdiagnosis and underreporting. These sources include ill persons who do not seek medical care, physicians who do not obtain stool specimens from patients with *E. coli* O157:H7 infection, laboratories that do not culture all stool samples for *E. coli* O157:H7, and the ability to detect antigen in *E. coli* O157:H7-contaminated stool samples (test sensitivity). However, before making this upward adjustment, the baseline annual number of cases was divided into two groups—cases with bloody diarrhea and cases with nonbloody diarrhea—by multiplying the baseline number of cases by the proportion expected to have bloody diarrhea (Table 4-2). Cases were divided into these two groups because the likelihood of seeking medical care, obtaining a stool specimen, and testing a stool specimen for *E. coli* O157:H7 is greater for patients with bloody diarrhea than for those with nonbloody diarrhea.

A negative binomial distribution was then applied to each of the sources of underdiagnosis and underreporting described above, providing an estimation of the number of missed cases. This procedure was completed separately for each of the two pathways: patients with bloody diarrhea and patients with nonbloody diarrhea. The negative binomial probability distribution outputs the number of failures (i.e., unreported cases), given inputs of the number of successes (reported cases) and the probability of success (estimates derived from the literature, described below). The probability of success used in the negative binomial probability distribution was estimated using a beta probability distribution. The output of the beta probability distribution is the prevalence (proportion) of an event, given inputs of the number of successes, s, and the number of trials, n.

The median (most likely) number of cases generated by the negative binomial distribution was used as the number of "missed cases" for each source of underdiagnosis/underreporting. Beginning with the baseline annual number of cases (described above), the missed cases for each source are summed for each group (those with bloody diarrhea and those with nonbloody diarrhea), and then the two group totals are summed to give an estimate of the total annual number of cases of symptomatic *E. coli* O157:H7 infection in the United States.

The data used as inputs into the negative binomial distribution are described above and summarized in Table 4-2. The distributions are listed in Table 4-3. The Monte Carlo simulation methods used here produce a distribution of possible values for each of the outcomes. Table 4-4 and Figure 4-2 show the results of this process, including estimates of the annual number of cases with bloody and nonbloody diarrhea and the total annual number of cases for the United States. The 2.5th and 97.5th percentiles represent the uncertainty about the number of cases at each step. The number of cases shown has been rounded to two significant digits to avoid

TABLE 4-2 Sources of Data Used to Estimate the Number of Undetected Cases of *E. coli* O157:H7 Infection

Event	Data	Reference(s)
Cases with bloody diarrhea are reported	640 of 757 (84.5%) patients with *E. coli* O157:H7 had bloody diarrhea	Ostroff et al. 1989 Hedberg et al. 1997 Slutsker et al. 1997 Kassenborg et al. 2001
Ill persons seek medical care	88 of 1,100 (8.0%) survey respondents reported seeking medical care for diarrhea	CDC 1998
	37 of 76 (48.7%) *E. coli* O157:H7 cases with bloody diarrhea sought medical care	Cieslak et al. 1997 Hedberg et al. 1997
Physicians obtain culture from patients	699 of 1,943 (36.0%) physicians surveyed obtain cultures from patients presenting with nonbloody diarrhea; 1,515 of 1,943 (78.0%) physicians obtain cultures from patients presenting with bloody diarrhea	Hedberg et al. 1997
Laboratories culture stool samples for *E. coli* O157:H7	108 of 230 (47.0%) labs surveyed test nonbloody stool for *E. coli* O157:H7	CDC 1997
	182 of 230 (79.1%) labs test bloody stool for *E. coli* O157:H7	
Sorbitol MacConkey agar (SMAC) test sensitivity	0.75 = probability a sample test is positive given it is infected	Hedberg et al. 1997

overstating the precision of the model. Outputs for intermediate steps in the pathway (e.g., probability of seeking care, probability of having a stool sample taken) are not shown.

Data Used to Adjust for Underdiagnosis and Underreporting

The proportion of patients who had bloody diarrhea was derived from the literature. A total of 640 (84.5%) of 757 reported cases of *E. coli* O157:H7 infection presented with bloody diarrhea (Ostroff et al. 1989; Hedberg et al. 1997; Slutsker et al. 1997; Kassenborg et al. 2001). These data were used in a beta distribution with inputs of the number of successes, s=640 (i.e., persons with bloody diarrhea), and the total sample size, n=757 (number of cases of symptomatic *E. coli* O157:H7 infection) (see Tables 4-2 and 4-3).

Information on the proportion of ill persons seeking medical care, physicians who obtain stool samples from symptomatic patients, and laboratory testing practices was obtained from FoodNet surveys and is summarized in Table 4-2. A high percentage of persons with bloody diarrhea seek medical care. Cieslak et al. (1997) found that 32 (55.2%) of 58 cases with bloody diarrhea in an *E. coli* O157:H7 outbreak in Las Vegas reported seeking medical care. Data from this and another study (Hedberg et al. 1997) were input into a beta distribution with inputs of s=37 and n=76 to determine the most likely number of missed cases at this step and the associated uncertainty. In the Hedberg et al. (1997) study, 88 (8.0%) of 1,100 respondents who had nonbloody diarrhea reported seeking medical attention. These values were used in a beta distribution with s=88 and n=1,100 (Tables 4-2 and 4-3).

TABLE 4-3 Input Values and Distributions Used to Estimate the Annual Number of Cases of *E. coli* O157:H7 Infection with Bloody and Nonbloody Diarrhea and the Total Number of Cases in the United States

Epidemiologic Parameter	Distribution	
Population-weighted, reported rate of *E. coli* O157:H7 per 100,000 person-years, all FoodNet sites, 1996–99	Discrete Uniform (1.53, 1.25, 1.95, 2.09)[a]	
U.S. population (1999)	272.7 million	
P(Case with bloody diarrhea is reported)[b]	Beta (640 + 1, 757 – 640 + 1)[c]	
P(Case with nonbloody diarrhea is reported)	1 – Beta (640 + 1, 757 – 640 + 1)	
	Bloody	Nonbloody
P(Laboratory cultures stool sample for *E. coli* O157:H7)	Beta (182 + 1, 230 – 182 + 1)	Beta (108 + 1, 230 – 108 + 1)
P(Physician obtains culture from patient)	Beta (1,515 + 1, 1,943 – 1,515 + 1)	Beta (699 + 1, 1,943 – 699 + 1)
P(Ill person seeks medical care)	Beta (37 + 1, 76 – 37 + 1)	Beta (88 + 1, 1,100 – 88 + 1)

[a]In a discrete uniform distribution, each of the four values listed in parentheses is equally likely to be sampled during simulations.

[b]*P*=probability of the event described.

[c]The input format for a beta distribution is (*s*+1,*n*–*s*+1), where *s*=the number of events of interest and *n*=total number of events measured (e.g., the number of cases with bloody diarrhea [*s*] and the number of all cases of symptomatic *E. coli* O157:H7 infection [*n*]).

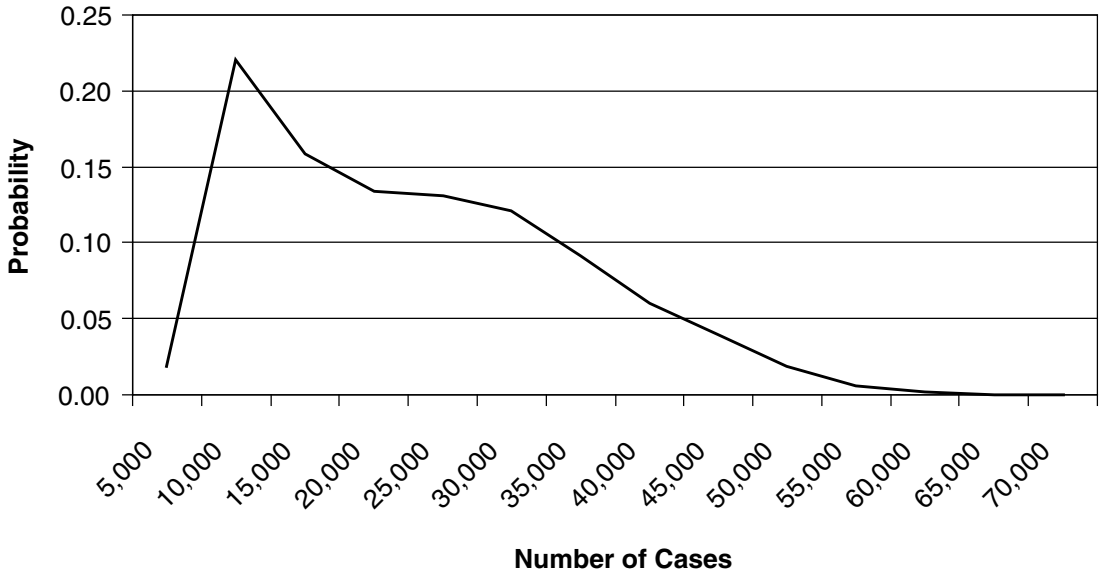

FIGURE 4-2 Estimated annual number of human cases of *E. coli* O157:H7 due to ground beef exposure.

TABLE 4-4 Number of Cases of Symptomatic *E. coli* O157:H7 Infection Due to All Exposures and Due to Exposure to Ground Beef Only (6,000 Iterations)

Number of Reported Cases	Median	2.5th and 97.5th Percentiles
All exposures		
Cases with bloody diarrhea	19,000	12,000 and 28,000
Cases with nonbloody diarrhea	74,000	45,000 and 116,000
Total annual cases	94,000	59,000 and 138,000
Ground beef exposures		
Cases with bloody diarrhea	3,800	1,000 and 9,000
Cases with nonbloody diarrhea	15,000	4,100 and 37,000
Total annual cases	19,000	5,300 and 45,000

Note: Number of cases has been rounded to two significant digits.

In a survey conducted in the FoodNet catchment area, 1,515 (78.0%) of 1,943 physicians reported that they obtained stool specimens from patients presenting with bloody diarrhea, and 699 (36.0%) of 1,943 physicians reported obtaining specimens from patients with nonbloody diarrhea (Hedberg et al. 1997). These data were fit to beta distributions (Tables 4-2 and 4-3).

In a national survey of clinical laboratories, 182 (79.1%) of 230 laboratories reported testing bloody stool for *E. coli* O157:H7 (CDC 1998), providing inputs for the beta distribution with s=182 and n=230 (Table 4-2). Only 108 (47.0%) of 230 laboratories reported testing all stool samples for *E. coli* O157:H7 (Hedberg et al. 1997), providing an s=108 and n=230 for input into a beta distribution (Tables 4-2 and 4-3). Hedberg et al. (1997) also reported that the sensitivity of the sorbitol MacConkey agar (SMAC) test used by the laboratories to identify *E. coli* O157:H7 in stool samples is 71% (Tables 4-2 and 4-3).

The model estimates that a median of 94,000 cases of symptomatic *E. coli* O157:H7 infection occur annually in the United States, accounting for underdiagnosis and underreporting (Table 4-4). Of these, an estimated 19,000 (20.2%) cases were characterized by bloody diarrhea. Mead et al. (1999) estimated that 73,480 cases of *E. coli* O157:H7 due to all exposures occur annually in the United States, approximately 20,000 fewer cases than the estimate derived by this risk assessment. This difference may be explained by differences in the methods used to derive the annual estimated number of cases. In the Mead et al. study, a weighted rate of 1.34 cases per 100,000 population was used to calculate a baseline number of cases. This rate is smaller than three of the four weighted rates used in this risk assessment (Table 4-1). In addition, Mead used a multiplier of 20 unreported cases for each reported case. In this risk assessment, unreported cases were estimated by several steps for two pathways: illnesses due to *E. coli* O157:H7 infection with bloody diarrhea and those with nonbloody diarrhea. This process resulted in 22 unreported cases for each reported case (94,000/4,200).

Estimating the Number of *E. coli* O157:H7 Illnesses Due to Contaminated Ground Beef (Etiologic Fraction)

To estimate the number of cases attributable to ground beef, the estimated total annual number of symptomatic *E. coli* O157:H7 infections for a given iteration was multiplied by an estimate of the proportion of cases due to exposure to ground beef (etiologic fraction). This calculation was done using Monte Carlo simulation and the inputs described below.

To estimate the etiologic fraction, data from studies of sporadic cases and outbreaks of *E.* coli O157:H7 were incorporated. Data from all outbreaks in which the route of transmission was identified were used, including those with waterborne and person-to-person transmission (CDC unpublished data). Two estimates were derived from outbreaks: the proportion of illnesses and the proportion of outbreaks due to ground beef exposure. During 1996 to 1999 (1999 is the most recent year for which data are available), ground beef was the most likely vehicle in 44 (30.1%) of 146 reported outbreaks of *E. coli* O157:H7 with an identified vehicle. This information was input into a beta distribution with s=44 and n=146. For the 146 outbreaks, 418 (11.1%) of 3,773 cases were attributed to ground beef; this information was input into a beta distribution with s=418 and n=3,773. Information from 48 outbreaks during 1996 to 1999 was excluded from this analysis.

Additional estimates of the etiologic fraction of illness due to ground beef contaminated with *E. coli* O157:H7 were obtained from four different case-control studies of mostly sporadic cases (MacDonald et al. 1988; Mead et al. 1997; Slutsker et al. 1998; Kassenborg et al. 2001). The etiologic fraction estimates for these studies were 17% (MacDonald et al. 1988), 26% (Mead et al. 1997), and 37% (Slutsker et al. 1998), and 7% and 8% for ground beef eaten away from home and at home, respectively (Kassenborg et al. 2001). The etiologic fractions of ground beef eaten at home or away from home were averaged to provide a single estimate.

These six estimates of the etiologic fraction, two from outbreak data and four from case-control studies, were input to a discrete distribution. Each of the six values were equally likely to be chosen during model simulation. During a given iteration, one of these six values was drawn at random. The etiologic fraction value drawn for a given iteration was multiplied by the estimated total number of cases for that iteration to arrive at the number of cases attributable to ground beef exposure. This process was repeated for the specified number of iterations during Monte Carlo simulation, producing a distribution of possible values for the annual number of cases attributable to ground beef.

Using the inputs described above, Monte Carlo modeling resulted in a median of 19,000 cases of symptomatic *E.* coli O157:H7 infection due to contaminated ground beef exposure (Table 4-4). However, uncertainty about the total number of cases implies that there may be fewer than 5,300 cases (2.5th percentile) or more than 45,000 cases (97.5th percentile) per year (Figure 4-2). This uncertainty distribution is used to develop the *E. coli* O157:H7 dose-response function. The median number of cases due to ground beef is 20.2% of the estimated median number of cases (94,000) due to all exposures.

DERIVING THE DOSE-RESPONSE FUNCTION FOR *E. COLI* O157:H7

The *E. coli* O157:H7 dose-response function was derived using information from three sources: (1) the estimated annual number of symptomatic *E. coli* O157:H7 infections due to ground beef exposure, (2) the estimated number of contaminated ground beef servings from the exposure assessment, and (3) the lower and upper bound dose-response curves derived using surrogate pathogens.

This section begins with a description of the beta-Poisson function used to fit dose-response data, followed by a description of how the lower and upper bound dose-response curves were developed from foodborne pathogens other than *E. coli* O157:H7 (surrogates). Then, the process for developing the dose-response function for *E. coli* O157:H7 in ground beef is described. The chapter ends with a discussion of the uncertainty about the estimated number of *E. coli* O157:H7 infections and contaminated servings.

Beta-Poisson Function

A beta-Poisson function was chosen to perform the dose-response analysis (Powell et al. 2000). This functional form assumes that a single organism is capable of infecting and inciting illness in an individual and that organisms operate independently within the host. Such assumptions are considered biologically plausible and defensible and can be used to derive a family of dose-response functions that include the beta-Poisson (WHO/FAO 2000; Buchanan et al. 2000; Haas et al. 1999).

Equation 4.1 is the beta-Poisson model, which predicts the probability of illness given a dose

$$p_i = 1 - (1 + d/\beta)^{-\alpha} \tag{4.1}$$

where

p_i = probability of illness,
α = alpha parameter,
d = dose of pathogen,
β = beta parameter = $N_{50}/(2^{\wedge}[1/\alpha] - 1)$, and
N_{50} = dose necessary to cause illness in 50% of those exposed.

The alpha and beta parameters needed in the beta-Poisson model are estimated by Equation 4.2, which is the maximum likelihood estimation routine developed by Regli et al. (1991). These estimates were obtained using the add-on program Solver, within Excel®. To briefly describe this process, Equation 4.2 is developed using an Excel spreadsheet, and the alpha and beta parameters are varied until Y is minimized. This process is performed separately for EPEC and *S. dysenteriae* data (described below):

$$\text{Y (minimized)} = 2\Sigma\{P_i * \ln(p_i/p_{oi}) + (T_i - P_i) * \ln[(1 - p_i)/(1 - p_{oi})]\} \tag{4.2}$$

P_i is the observed number of positive responses at the ith dose, p_{oi} is the observed proportion of response at the ith dose, T_i is the total number of subjects in the ith dose group, and p_i is the response estimated by the beta-Poisson function at the ith dose.

The output of these beta-Poisson models is the estimated proportion of persons expected to experience illness given a dose. The proportion of persons expected to fall ill at a given dose multiplied by the number of servings containing that dose, as estimated by the exposure assessment portion of the model, results in an estimate of the number of persons expected to become ill during a year.

Developing Upper and Lower Boundaries to the *E. coli* O157:H7 Dose-Response Function

No human clinical trial data are available for *E. coli* O157:H7, but they are available for a number of pathogens that can be used as surrogates (see Appendix B). These surrogates are used to form upper and lower boundaries between which the *E. coli* O157:H7 dose-response function is assumed to fit. This method is termed the envelope method because these upper and lower boundaries envelop the *E. coli* O157:H7 dose-response function (Vose 1996, p. 202). Therefore, the upper and lower boundaries describe the extent of uncertainty about the true *E. coli* O157:H7 dose-response.

Several *Shigella* and other *E. coli* species were considered as possible surrogates. In considering a species to use as a surrogate, a number of factors were evaluated, including availability of data, genetic relatedness, and similarities in transmission, infectivity, and pathogenicity. Other risk assessments of *E. coli* O157:H7 have used *Shigella* as a surrogate pathogen (Cassin et al. 1998; Marks et al. 1998).

E. coli O157:H7 may be most similar to *Shigella* spp. with regard to transmission and infectivity; however, *Shigella* spp. are invasive pathogens that multiply within host epithelial cells, whereas *E. coli* O157:H7 does not. Both are transmitted by food, although humans are the reservoir of *Shigella* spp. contamination of food and water. The probability of infection with low doses of *Shigella* spp. is thought to be high. There are four species of *Shigella* spp.: *S. sonnei*, *S. flexneri*, *S. boydii*, and *S. dysenteriae.* A clinical experiment in human volunteers has been conducted using *S. sonnei*; however, this trial used only one dose of pathogen. Without multiple data points in the form of administered dose levels, a curve cannot be fitted to generate parameters for the dose-response function; therefore, *S. sonnei* was not used as a surrogate. A substantial amount of human experimental data are available for one strain of *S. flexneri*; however, this organism does not produce Shiga toxins and thus was not chosen as a surrogate.

S. dysenteriae was selected as an upper bound to the *E. coli* O157:H7 dose-response function based on the assumption that *E. coli* O157:H7 is unlikely to be more pathogenic than this invasive *Shigella* species. Both *S. dysenteriae* type 1 and *E. coli* O157:H7 strains produce Shiga toxins, a virulence factor that appears to increase the severity but not necessarily the probability or frequency of illness. Similar to *E. coli* O157:H7, *S. dysenteriae* has a high probability of illness associated with low doses; both organisms cause hemolytic uremic syndrome (HUS).

The data for *S. dysenteriae* (Table 4-5) include dose groups of 4, 6, or 10 volunteers administered four-dose levels from 10 to 10,000 pathogen cells (Levine et al. 1973). These trials found a generally increasing proportion of symptomatic infection as the dose was increased, with 10% of persons exposed at the lowest doses and 83% of those exposed at the highest doses developing clinical symptoms.

Another surrogate, enteropathogenic *E. coli* (EPEC), was chosen to represent the lower bound of an *E. coli* O157:H7 dose-response function, based on the assumption that *E. coli* O157:H7 is unlikely to be less pathogenic than the EPEC. EPEC and *E. coli* O157:H7 have similar mechanisms of transmission, that is, by food, water, and person-to-person contact; however, unlike *E. coli* O157:H7, EPEC is principally a disease of children younger than 1 year of age and generally requires large doses (e.g., 100 million organisms) before a substantial probability of illness is observed. A substantial amount of data were available from human clinical trials for EPEC (Levine et al. 1978; Bieber et al. 1998). Therefore, three virulent EPEC strains were selected as surrogates. The data for EPECs (Table 4-6) include dose groups of two, four, five, or six volunteers administered six-dose levels from 10^6 to 10^{10} pathogen cells. In some trials, no one developed symptomatic infection at lower doses; all persons developed symptomatic infection at higher doses.

The dose and response information found in Tables 4-5 and 4-6 was used in Equations 4.1 and 4.2. Dose-response calculations were performed separately for each of the two surrogate organisms.

The estimated lower bound dose-response generated using EPEC clinical trial data and the estimated upper bound dose-response generated using *Shigella dysenteriae* data are illustrated in Figure 4-3. The estimated alpha and beta parameters are shown in Table 4-7.

TABLE 4-5 Data from Human Volunteers Administered Two Strains of *Shigella dysenteriae*

Shigella Dysenteriae Strain	Dose of Pathogen	Number of Persons Developing Symptoms	Total Persons Exposed	Proportion of Persons Developing Symptoms
M 131	10	1	10	0.10
A-1	200	1	4	0.25
M 131	200	2	4	0.50
M 131	2,000	7	10	0.70
A-1	10,000	2	6	0.33
M 131	10,000	5	6	0.83

Source: Levine et al. 1973.

TABLE 4-6 Data from Human Volunteers Administered Four Strains of Enteropathogenic *Escherichia coli* (EPEC)

EPEC Strain	Dose of Pathogen	Number of Persons Developing Symptoms	Total Persons Exposed	Proportion of Persons Developing Symptoms
O128	1,000,000	0	5	0.00
O127	1,000,000	0	4	0.00
O142	1,000,000	1	5	0.20
O128	100,000,000	0	5	0.00
O142	100,000,000	1	5	0.20
B-171-8	500,000,000	3	5	0.6
B-171-8	2,500,000,000	6	6	1
O128	10,000,000,000	0	5	0.00
O127	10,000,000,000	3	5	0.60
O142	10,000,000,000	5	5	1.00
B-171-8	20,000,000,000	2	2	1

Sources: Bieber et al. 1998; Levine et al. 1978.

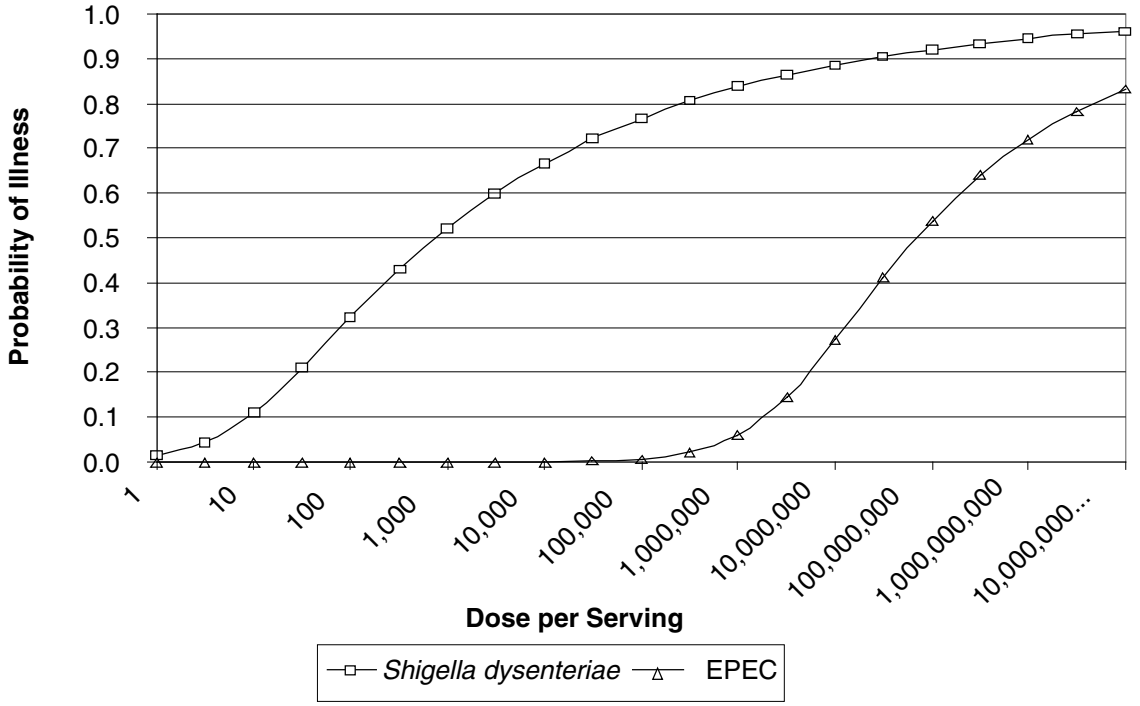

FIGURE 4-3 Dose-response curves for *Shigella dysenteriae* (Shig dys cumulative distribution function [cdf]) and enteropathogenic *E. coli* (EPEC cdf). The *Shigella dysenteriae* curve serves as the upper bound and EPEC as the lower bound to a dose-response curve for *E. coli* O157:H7.

TABLE 4-7 Alpha and Beta Parameters for the Upper and Lower Bound Beta-Poisson Models, *Shigella dysenteriae* and Enteropathogenic *Escherichia coli* (EPEC)

Surrogate Organism	Alpha	Beta
S. dysenteriae	0.157	9.17
EPEC	0.221	3,110,000

For the EPEC dose-response curve, the implied dose at which 50% of persons exposed will become ill (N_{50}) is 68 million organisms. *E. coli* O157:H7 is highly unlikely to have an N_{50} that is this high. For the *Shigella dysenteriae* dose-response curve, the N_{50} is 740 organisms. By using this pathogen to represent an upper bound, it is assumed that *E. coli* O157:H7 is unlikely to have an N_{50} lower than 740 organisms.

Process for Developing the *E. coli* O157:H7 Dose-Response Function

A beta-Poisson dose-response function for *E. coli* O157:H7 is derived from the distribution of *E. coli* O157:H7 illnesses attributable to ground beef (response) and the distribution of the number of *E. coli* O157:H7 organisms in consumed ground beef servings (dose) (Powell et al. 2000). The derived *E. coli* O157:H7 dose-response function is constrained to lie between beta-Poisson functions fit to *Shigella dysenteriae* and EPEC data. The derivation can be simply represented as

$$\text{Ex} \times \text{DR} = \text{TC} \tag{4.3}$$

where Ex represents the exposure distribution, DR is the dose-response function, and TC is the total cases per year. If dose-response data for *E. coli* O157:H7 were available, a dose-response function could be fit to these data and Equation 4.3 could be directly solved for total cases. In the absence of dose and response data, available estimates for the total number of cases can be used with the model's estimates for the exposure distribution to determine the dose-response using Equation 4.3.

Uncertainty in Cases and Exposure Distribution

Uncertainty about the exposure distribution predicted by this model was illustrated as an output from Chapter 3. Uncertainty about the number of *E. coli* O157:H7 cases associated with ground beef was discussed above. These uncertainties are integrated in deriving the dose-response function for *E. coli* O157:H7 (Figure 4-4).

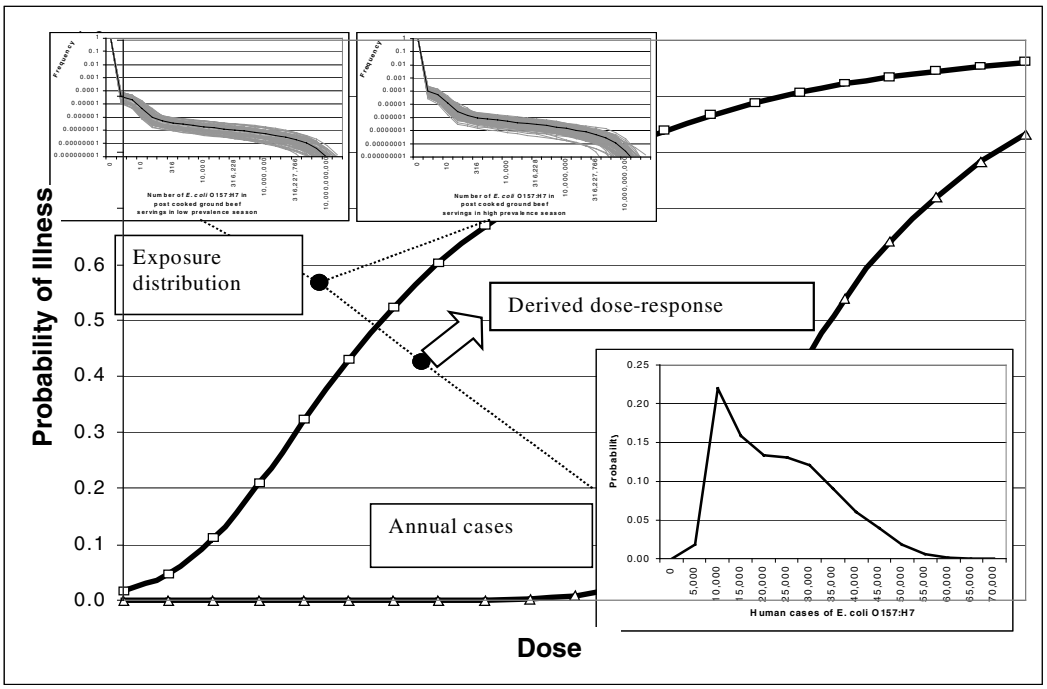

FIGURE 4-4 Illustration of process by which an *E. coli* O157:H7 dose-response function is derived from uncertain exposure distributions and uncertain total human cases.

Because the exposure assessment estimates exposure distributions for high and low prevalence seasons but the estimated total cases distribution reflects an annual number, the two seasonal exposure distributions must be combined to represent exposures in servings of ground beef across a full year. This annual exposure distribution is estimated by weighting each seasonal distribution by its number of months. Therefore, the high prevalence season is given a weight of 4 ÷ 12 (for June through September) and the low prevalence season is given a weight of 8 ÷ 12.

For a given draw from the uncertain exposure distributions and the total cases distribution, a best-fitting beta-Poisson function is determined by varying the alpha and beta parameters of that function. These parameters are constrained, however, by the lower and upper bound parameters estimated for *Shigella dysenteriae* and EPEC. The set of parameter values that result in predicting the specified total cases of *E. coli* O157:H7 illness given the exposure distribution is

saved, and the algorithm is repeated for another draw from the exposure and total cases distributions. With this method, the uncertainty regarding exposures and total cases per year is fully integrated into the estimate of a dose-response function.

Figure 4-5 shows the resulting uncertainty about the derived *E. coli* O157:H7 dose-response function. Each curve shows progressively higher percentiles of the derived dose-response function extending from the 5th to the 95th percentiles. The median dose-response function in this range is assumed to be the best estimate.

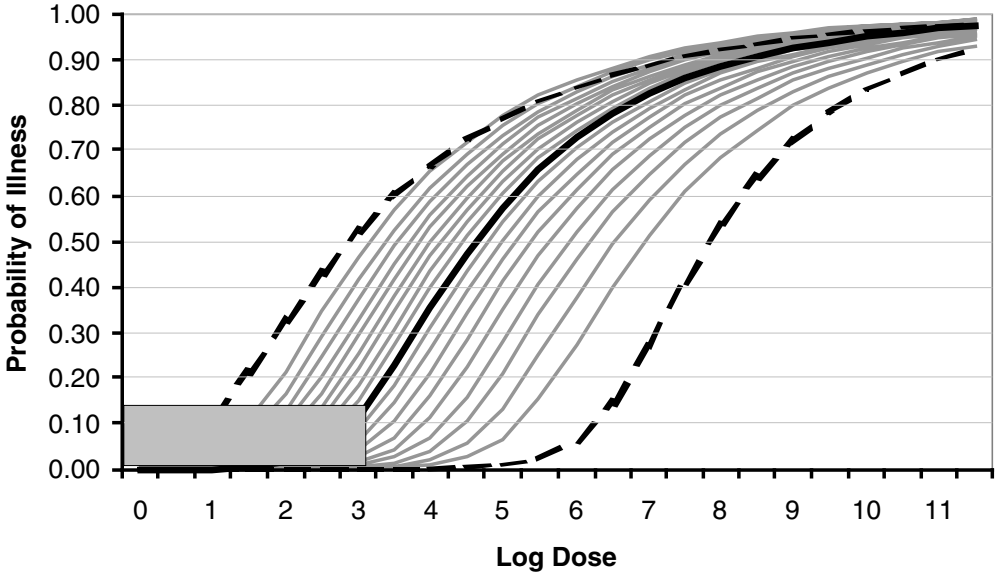

FIGURE 4-5 Derived dose-response curves from combining output of hazard characterization and exposure assessment. Curves represent percentiles of uncertainty distribution (ranging from 5th to 95th percentile) about the *E. coli* O157:H7 dose-response function. The thick line is the median dose-response curve. The dashed lines are boundary dose-response functions fit to *Shigella dysenteriae* and enteropathogenic *E. coli* (EPEC). The rectangle in the lower left represents the combined range of uncertainty of the dose and response derived from the 1994 outbreak in the northwestern United States. Source: Bell et al. 1994.

Uncertainty about the *E. coli* O157:H7 dose-response function extends almost across the full range enveloped by the lower and upper bound curves. Nevertheless, this uncertainty suggests more confidence in dose-response functions that lie closer to the *Shigella dysenteriae* boundary than in those that lie closer to the EPEC boundary. Therefore, the results of this derivation suggest that the dose-response function for *E. coli* O157:H7 more closely approximates that estimated for *Shigella dysenteriae* than for EPEC.

The derived dose-response function for *E. coli* O157:H7 also shows consistency with information obtained from a ground beef-associated outbreak in the northwestern United States. Uncertainty about the average exposure dose and attack rate in this outbreak is shown in Figure 4-5. The majority of the percentiles for the derived dose-response function fit within the outbreak's uncertainty range. However, even the boundary formed by the *Shigella dysenteriae* dose-response function fails to explain all of the outbreak's uncertainty.

Effect of Uncertainty in Exposures and Cases

Although the derivation of the *E. coli* O157:H7 dose-response function includes the uncertainty from the exposure assessment and the total number of cases occurring per year, it does not suggest the relative contribution of each source of uncertainty to the overall uncertainty in the dose-response function. To examine this relative effect, the exposure distribution and the total cases were considered fixed, in turn, and the uncertainty about the other was used to derive the dose-response function.

Figure 4-6 shows dose-response functions estimated by setting the exposure distribution at its median but using the 5th and 95th percentiles from the total cases per year distribution. The N_{50} for the dose-response curve fit to the 5th percentile of cases is about 3.5 logs. In other words, the dose-response function predicts that 50% of those exposed to an average dose of 3.5 logs of *E. coli* O157:H7 will become ill. The N_{50} for the dose-response curve fit to the 95th percentile of cases is about 6.5 logs. This range in uncertainty is slightly less than the range shown in Figure 4-5. It is also reasonably symmetrical about the median curve shown in Figure 4-5.

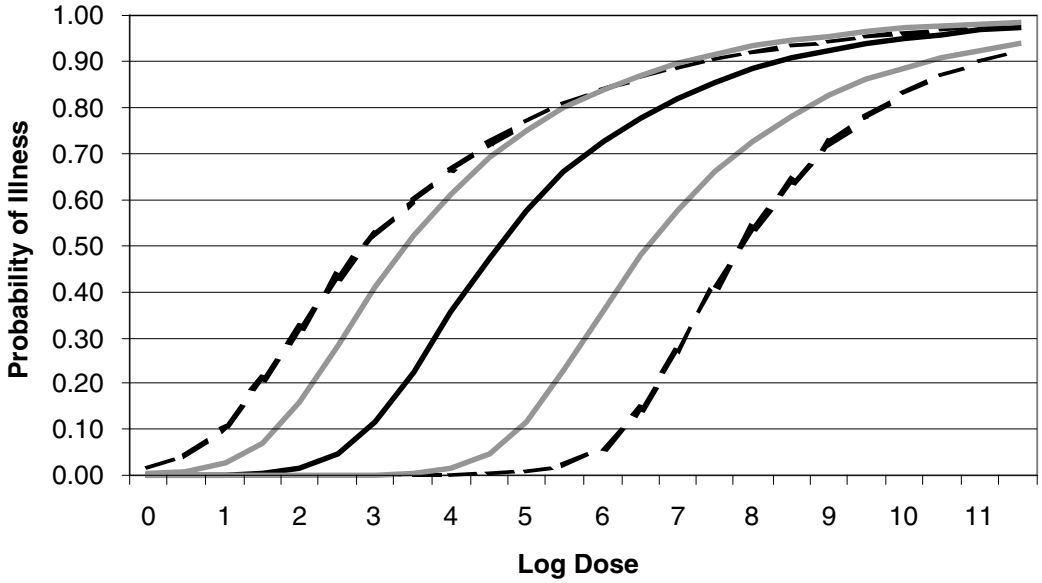

FIGURE 4-6 Dose-response curves that result from setting exposure distribution at the median and using 5th and 95th percentiles (grey lines) of cases predicted from hazard characterization. The solid dark line is the median dose-response function including uncertainty about exposures and cases (Figure 4-5). The dashed lines are boundary dose-response functions fit to *Shigella dysenteriae* and enteropathogenic *E. coli* (EPEC).

Figure 4-7 shows dose-response functions estimated by setting the total number of cases at the median but using the 5th and 95th percentiles from the exposure distribution. This figure is generally similar to Figure 4-6. In Figure 4-7, the N_{50} for the dose-response curve fit to the 5th percentile of the exposure distribution is about 3 logs. The N_{50} for the dose-response curve fit to the 95th percentile of the exposure distribution is about 6 logs. This range is shifted to the left relative to the range shown for Figure 4-6. This shift implies that fixing the number of cases at its median value would move estimated dose-response functions closer to the *Shigella dysenteriae* curve than fixing the exposure distribution at its median (as done in Figure 4-6).

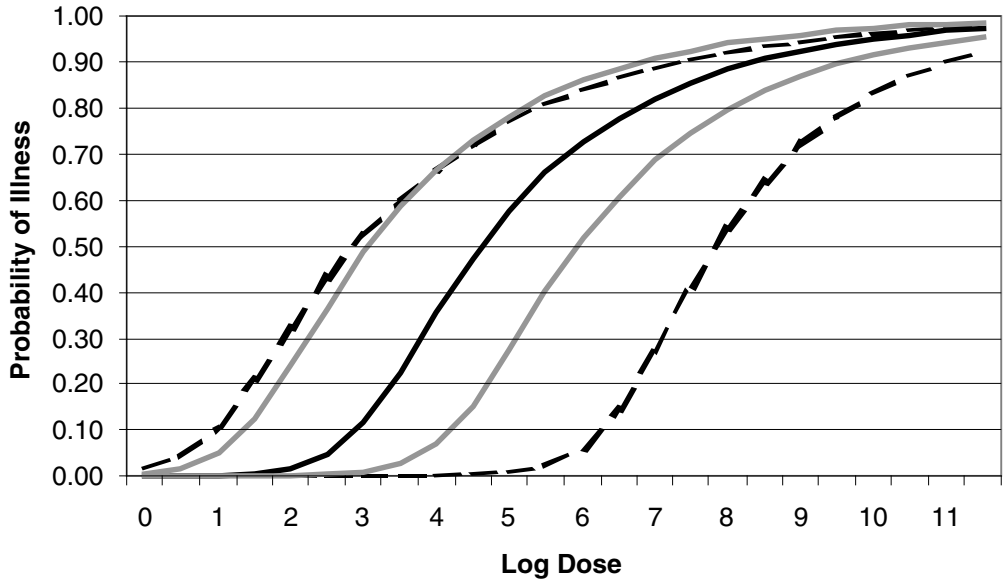

FIGURE 4-7 Dose-response curves that result from setting total *E. coli* O157:H7 cases per year at the median and using 5th and 95th percentiles (grey lines) of the exposure distribution predicted from the exposure assessment. The solid dark line is the median dose-response function including uncertainty about exposures and cases (Figure 4-5). The dashed lines are boundary dose-response functions fit to *Shigella dysenteriae* and enteropathogenic *E. coli* (EPEC).

The implication of this analysis is that neither uncertainty about the exposure distribution nor uncertainty about the total number of cases dominates the uncertainty about the *E. coli* O157:H7 dose-response function. Instead, both sources of uncertainty contribute equally to the overall uncertainty.

ESTIMATING SEVERE CLINICAL OUTCOMES DUE TO *E. COLI* O157:H7 INFECTION

The estimates generated by this portion of the model are not used in developing a dose-response curve for *E. coli* O157:H7. Instead, they describe the consequences of symptomatic infection. Given the lack of dose-response data, the probability of various clinical outcomes is assumed to be independent of the dose of *E. coli* O157:H7 consumed. Estimating the clinical outcomes of symptomatic infection is essential for future cost-benefit analyses of intervention options. Estimates are provided for severe illnesses due to ground beef exposure and due to all exposures.

The number of persons who experienced hospitalization, HUS, thrombotic thrombocytopenic purpura (TTP), or death was estimated using data from 203 outbreaks that occurred between 1982 and 1998 (CDC unpublished data). A total of 4,478 cases occurred during the 203 outbreaks; of these, 968 (21.6%) cases resulted in hospitalization, 228 (5.1%) cases progressed to HUS or TTP, and 28 (0.6%) cases resulted in death (Table 4-5). Only summary data were available for these outbreaks, preventing calculation of conditional probabilities. Therefore, for the purposes of modeling, it is assumed that HUS or TTP cases occur only among hospitalized patients and that deaths occur only among those patients with HUS or TTP. The data from these outbreaks were used as inputs to beta distributions and simulated.

The data inputs estimating the number of hospitalizations were *s*=968 and *n*=4,478; the number of cases with HUS or TTP, *s*=228 and *n*=968; and the number of deaths, *s*=28 and *n*=228. Recall that *s* is the number of events of interest and *n* is the total number observed. Applying these proportions to all cases assumes that pathogenicity is similar among strains of *E. coli* O157:H7 and that outcome is independent of dose.

The median annual estimated number of patients with bloody diarrhea who sought medical care was 9,400 (Table 4-8); of these, 2,000 (21.3%) persons were hospitalized. Of the hospitalized patients, the model estimates that a median of 460 (23.0%) patients developed HUS or TTP and that 50 (10.9%) of these patients died. These estimates are similar to the estimated 2,168 hospitalizations and 52 deaths annually due to *E. coli* O157:H7 infection reported by Mead et al. (1999).

The proportion of cases with severe clinical outcomes attributable to ground beef exposure is also presented in Table 4-8. The model estimates that a median of 1,800 severe cases (patients with bloody diarrhea who sought medical care) are due to ground beef exposure annually. Of these 1,800 cases, the model estimates that 400 (22.2%) will be hospitalized and that, of these, 90 (22.5%) will develop HUS or TTP and 10 (11.1%) HUS/TTP patients will die.

TABLE 4-8 Number of Severe Outcomes Due to *E. coli* O157:H7 Infection and the Distributions and Inputs Used to Calculate These Outcomes (6,000 Iterations)

Parameter	Distribution	
Proportion of cases hospitalized	Beta (968 + 1, 4,478 – 968 + 1)[a]	
Proportion of hospitalized cases progressing to HUS/TTP	Beta (228 + 1, 968 – 228 + 1)	
Proportion of HUS/TTP cases resulting in death	Beta (28 + 1, 228 – 28 + 1)	
Severe Health Outcomes	**Median**	**2.5th and 97.5th Percentiles**
All exposures		
Severe (patient with bloody diarrhea, seeks medical care)	9,400	6,300 and 12,000
Hospitalized	2,000	1,300 and 2,600
HUS/TTP	460	300 and 630
Deaths	50	30 and 100
Ground beef exposures		
Severe (patient with bloody diarrhea, seeks medical care)	1,800	1,000 and 4,100
Hospitalized	400	100 and 900
HUS/TTP	90	30 and 210
Deaths	10	1 and 30

Note: Number of cases has been rounded to two significant digits (one significant digit for numbers less than 100). HUS = hemolytic uremic syndrome; TTP = thrombotic thrombocytopenic purpura.

[a]The input format for a beta distribution is (*s*+1,*n*–*s*+1), where *s*=the number of events of interest and *n*=total number of events measured (e.g., the number of cases with bloody diarrhea [*s*] and the number of all cases of symptomatic *E. coli* O157:H7 infection [*n*]).

SENSITIVE SUBPOPULATIONS

Certain age groups have a higher reported incidence of *E. coli* O157:H7 infection. Surveillance from FoodNet sites in 1999 shows that 1- to 9-year-olds had the highest incidence among all age groups (Figure 4-8, CDC 2000). Nationwide in 1998, 1- to 4-year-olds had the highest incidence, at 4.57 reported cases per 100,000 population (CDC 1999). Young children also appear to be more susceptible to developing HUS (see Chapter 2).

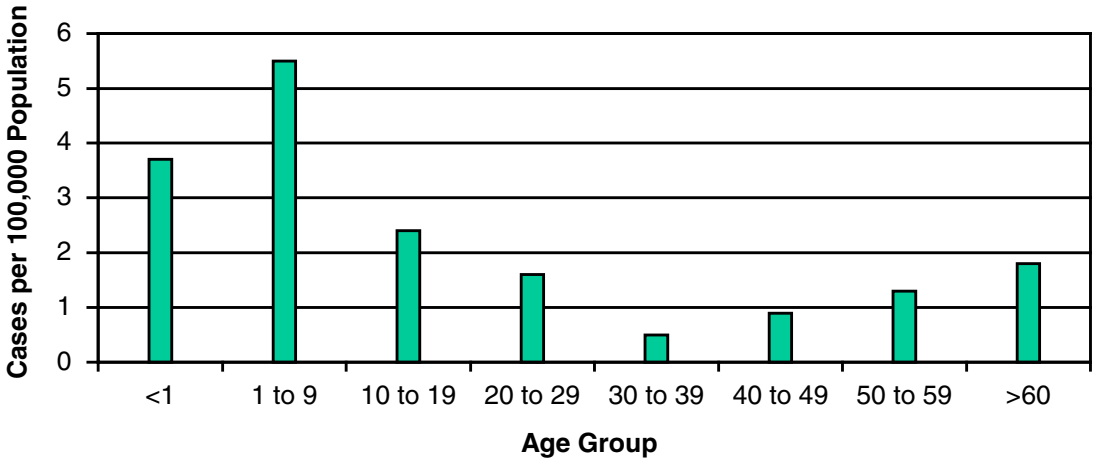

FIGURE 4-8 Number of reported cases of *E. coli* O157:H7 infection due to all routes of transmission, by age group, FoodNet sites, 1999.

The reason why children have the highest reported incidence of *E. coli* O157:H7 infection is not known. Relative to adults, children may be more likely to receive medical care during an episode of diarrhea or bloody diarrhea and be more likely to be tested for *E. coli* O157:H7. They may also have better access to health care, a higher likelihood of being reported to public health officials, more opportunities for exposure, increased susceptibility to infection, or some combination of all of these factors. Children are more likely than adults to develop HUS as a sequela of infection with *E. coli* O157:H7. Kidney damage that occurs during HUS is a result of Shiga toxin binding to specific receptors present on kidney cells. These receptors appear to be present in the kidneys of children but not adults (Lingwood et al. 1998).

Given that children consistently have the highest rate of *E. coli* O157:H7 infection relative to older age groups, it would seem reasonable to conduct a separate dose-response analysis of children. However, data on the proportion of *E. coli* O157:H7 infections due to ground beef, by age, are scarce. Also, the epidemiology of *E. coli* O157:H7 in children is complex, as described above. It is not known whether the high incidence in children is due to more children having the disease relative to adults or to artifacts of the health care and public health reporting systems, and little or no data are available to answer these questions. Therefore, this risk assessment does *not* include a separate dose-response analysis for children.

VALIDATION OF THE *E. COLI* O157:H7 DOSE-RESPONSE FUNCTION USING OUTBREAK DATA

An epidemiologic investigation traced a 1992 to 1993 outbreak of *E. coli* O157:H7 to consumption of hamburgers at a chain of fast-food restaurants (Chain A) in the Pacific

Northwest (Bell et al. 1994). Data from this investigation were used to develop a separate dose-response function to validate the dose-response function derived separately in this risk assessment. The data were not used directly in this risk assessment because the contamination levels of *E. coli* O157:H7 in the ground beef servings were not directly correlated with the severity of illness (i.e., the number of *E. coli* O157:H7 organisms consumed by each human case was not known).

A total of 501 culture-confirmed cases were documented to occur during this outbreak, including 398 (79.4%) primary cases, 48 (9.6%) secondary cases, and 55 (11.0%) cases that could not be classified as either primary or secondary. Of the 398 patients with primary disease in Washington state, 374 (93.9%) had eaten at Chain A in the previous 10 days. A total of 344 (92.0%) of 374 primary cases who ate at Chain A reported eating a regular (45-gram) hamburger. The median age of cases was 8 years, ranging from 4 months to 88 years. Forty-five (9.0%) patients developed HUS and 3 died of complications of HUS. The median age of HUS patients was 5 years, ranging from 1 to 68 years.

In response to the outbreak, approximately 255,000 45-gram hamburger patties were recalled from Chain A restaurants in Washington (Bell et al. 1994). These patties had been produced on November 19, 1992, at a processing plant in California. The recalled patties represented 43% of all regular hamburgers produced for Chain A at the California plant on that day, for a total production of 593,023 patties. The processing plant had sent 62% of that day's production (367,673 patties) to Washington. Therefore, the number of hamburger patties sold and consumed was equal to the number sent to Washington minus the number recalled (367,673 – 255,000 patties), or 112,673 patties.

The number of *E. coli* O157:H7 organisms per serving that occurred during this outbreak was quantified in six raw ground beef samples from implicated lots (Marks et al. 1998). The samples were enumerated for *E. coli* O157:H7 by the most probable number (MPN) method and were found to contain 0.3, 0.9, 1.5, 2.8, 4.3, and 15 colony-forming units per gram (CFU/g), respectively (Johnson et al. 1995; Tuttle et al. 1999). The distribution for the concentration of *E. coli* O157:H7 in the raw ground beef, d, was modeled by assuming that the quantity

$$\{[\ln(d)] - m\}/[s(1/n)^{1/2}] \sim t_{n-1}$$

is distributed as a t-distribution with $n-1$ degrees of freedom, where m is the mean of the $n=6$ log densities and s is the standard deviation of the log densities. Multiplying the distribution for d (CFU/g) by a serving size of 45 grams yields an estimated median per serving load of 96 CFU of *E. coli* O157:H7 before cooking (90% confidence interval, 5 to 1,844 CFU). This estimate is similar to the findings of Tuttle et al. (1999), who calculated a median of 67.5 organisms per raw ground beef patty (range, 13.5 to 675 organisms per patty).

To determine the effect of cooking on the final number of *E. coli* O157:H7 organisms, Bell et al. (1994) reported cooking 16 regular hamburgers according to Chain A's routine practices. After the frozen patties were cooked for 1 minute on each side on a 191°F grill, all of them had at least one internal temperature measurement below 68.3°C (155.0°F) (range, 41.7 to 81.1°C [107.1 to 177.98°F]). Ten had a measurement below 60.0°C (140°F). The minimum internal cooking temperature was modeled as a custom cumulative distribution with a minimum value of 37.8°C (100°F), a 6.25th percentile of 41.7°C (107.1°F), a 62.5th percentile of 60.0°C (140.0°F), and a maximum of 68.3°C (155.0°F).

Based on a study by Juneja et al. (1997), Marks et al. (1998) predicted the log reduction of *E. coli* O157:H7 in hamburgers due to cooking to be

$$\log_{10} (N_f/N_0) = 13.93 - 0.12*T$$

where N_0 is the number of organisms before cooking, N_f is the number of organisms after cooking, and T is cooking temperature (°F). Combining the distributions for the number of organisms in a raw patty prior to cooking with the cooking temperatures described above, this equation suggests that cooking rendered 50% of the hamburger patties free of *E. coli* O157:H7. From the estimate described above, 112,674 patties were purchased at Chain A restaurants; therefore, half of these, or 56,337 patties, were estimated to still be contaminated after cooking. The simulated distribution for the amount of viable *E. coli* O157:H7 per serving that remained after cooking has a median value of 23 CFU per serving (1 and 926 CFU per serving, 2.5th and 97.5th percentiles, respectively). This simulated estimated number of organisms is in agreement with a study of 76 recalled ground beef patties from this outbreak (Tuttle et al. 1999), where the median most probable number of organisms was determined to be 67.5 per uncooked patty (range, 13.5 to 675).

At this ingested dose, the uncertainty about the attack rate is estimated using a beta distribution. Inputs to this distribution were the number of primary cases (374) that had eaten at Chain A in the 10 days prior to illness, adjusted for underdiagnosis and underreporting (the input, *s*), and the number of patties contaminated with at least one *E. coli* O157:H7 organism after cooking (*n*=56,337). To adjust for underdiagnosis and underreporting, the number of primary cases that had eaten at Chain A was multiplied by a factor of 1 to 20 using a uniform probability distribution. A uniform distribution randomly chooses a value in the specified range during a given iteration. Therefore, 374 was multiplied by the randomly drawn value between 1 and 20 during each iteration of the model, resulting in a list of the possible number of actual cases that had occurred during the outbreak.

Modeling the underreporting factor in this manner accounts for uncertainty in the degree of underreporting that had occurred during this outbreak. A factor of 1 indicates no underreporting occurred; a factor of 20 indicates that 20 cases occurred for each reported case and is the underreporting factor used in Mead et al. (1999) for *E. coli* O157:H7. Because of the extensive publicity about this outbreak, the degree of underreporting is likely to be somewhat less than the estimated national average of 20.

A Monte Carlo simulation of 100,000 iterations resulted in a median value of 70 cases per 1,000 contaminated servings consumed (10 and 130 cases per 1,000 servings, 2.5th and 97.5th percentiles, respectively) at a median of 23 CFU per serving. For the outbreak, the probability of illness given a dose is consistent with the *E. coli* O157:H7 dose-response curve in Figure 4-5. In this figure, the outbreak information is represented by the rectangle in the lower left corner of the graph.

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